

ON THE RESPIRATION IN SCALLOPS (LAMELLIBRANCHIATA)¹

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The only data in the literature on the respiration of scallops seem to be those of Späreck (1936; cf. also Thorson, 1936), who determined the oxygen uptake in *Pecten groenlandicus* (arctic), *P. varius* (boreal-mediterranean), and *P. flexuosus* (mediterranean) in nearly air-saturated water. This author found that the resting metabolism of the scallops, and other bivalves which have a marked capacity for rapid swimming locomotion, is considerably higher than that of more sedentary types.

The present paper reports on a study of the respiration in the deep sea scallop and the bay scallop, both of common occurrence and of considerable economic importance along certain areas of the Atlantic coast of North America. The oxygen consumption of resting (non-swimming) individuals was determined over a range of concentrations of oxygen of about the air-saturation value to close to zero. In addition determinations were made, in aerated water, of the oxygen utilization or the percentage of oxygen removed from the inspired water during its passage through mantle cavity and gills.

MATERIAL AND METHOD OF OXYGEN ANALYSIS

The deep sea scallops (*Pecten grandis* Solander) were taken from Cape Cod Bay, at a depth of about 12 m., the bay scallops (*Pecten irradians* Lamarck) from a shallow salt water pond near Woods Hole. All animals were kept in the laboratory in tanks with running sea water (salinity 31–32.5‰). For the experiments, which were carried out during June and July, 1952, and from September–November, 1953, only specimens that were in apparent good health, showing a wealth of actively moving, extended tentacles and production of faecal material, were used.

Analyses of dissolved oxygen were made by the Winkler method on samples of about one ml. (van Dam, 1935a).

TECHNIQUES OF MEASUREMENTS OF O₂-CONSUMPTION

(a) Continuous measurements of O₂-consumption in water approximately saturated with air were performed on single individuals of the sea scallop by means of a volumetric respirometer technique as described by Scholander (1949). In this method the oxygen consumed is directly read on a syringe from which oxygen is introduced into the system at the rate at which it is removed by the animal.

A plastic box of about 900 ml. capacity, immersed in a water bath, was used as a respiration chamber. It was filled with about 850 ml. of sea water which had been filtered through cotton or glass wool. Prior to the actual experiments the animals were kept for one hour or longer in the respirometer running open to the

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air. Runs without an animal in the water showed a blank oxygen consumption of, on the average, about 0.2 ml. O_2 /hr. (Fig. 5). The cause of these rather high blank values is not known. The calculated Q_{O_2} -values (O_2 -consumption per kg. and hour) given in Figure 4 are STP- and blank-corrected. In a number of tests initial and final O_2 -content and approximate pH (Hellige comparator) of the water in the respirometer were determined, showing that, as a rule, the O_2 -content remained practically constant. In a few cases a decrease of 0.1–0.3 ml. O_2 /L. was observed, and the Q_{O_2} -values were corrected accordingly. Although in this method the CO_2 produced is continuously being removed, the pH of the water during the tests usually decreased from about 8 to about 7.2–7.6. This, however, as can be seen from Figure 5, had no apparent effect on the rate of oxygen uptake. The same holds for the experiments with the closed respiratory chamber technique described below, in which all excreta, CO_2 included, remained in the water.

(b) To determine the critical O_2 -tension, measurements of O_2 -consumption were made in closed plastic respiration chambers of 1–3 L. capacity (Fig. 1), completely

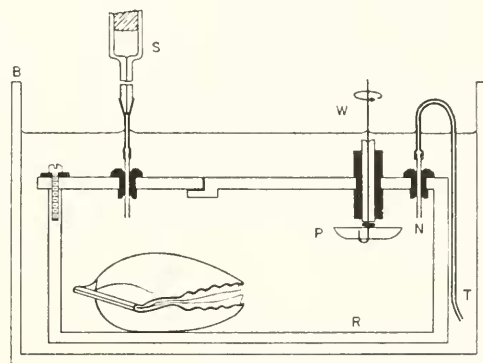


FIGURE 1. Diagram of closed respiratory chamber (R). W, P: stainless steel drive shaft and plastic paddle. T, N: small bore plastic tubing and needle through which water from water-bath B enters R when water samples for O_2 -analysis are taken with syringe S.

filled with water, in which the concentration of oxygen decreased progressively due to the respiration of the animal.

Blank runs (Fig. 6) showed that in the absence of an experimental animal the O_2 -content of the water in the respirometer was practically constant at any O_2 -tension. Control tests showed that the Winkler procedure, as such, was not interfered with by metabolites in water in which animals had been confined for several hours.

Prior to the O_2 -uptake measurements the animals were scrubbed clean thoroughly and acclimated to the experimental temperatures for one day or longer. To determine the wet weight of the soft tissues, the animal was opened by severing the adductor muscle. Then all soft tissues were taken out of the shells, drained for about 5 minutes, dried at the outside on filter paper and then weighed. Length and weight of the animals used are given in Table I.

TECHNIQUE OF DETERMINING THE OXYGEN UTILIZATION

The sea scallops were lying freely in a normal position, *i.e.*, on the right valve, on the bottom of an all-glass aquarium. Samples of the inhalant water current were taken with a syringe, the tip of which was placed near the pallial opening opposite the middle of the hinge. Simultaneously, samples of the exhalant current were taken, either directly by means of a syringe to which a fine glass tip, bent at right angles, was attached (animals no. 1 and 5, 18 samples), or by means of a siphon as

TABLE I
*Length and weight of experimental animals**

Specimen no.	Number of test in which specimen was used		Length (mm.)	Wet meat (gms.)	Total wet weight (gms.)
	Test no.	Fig. no.			
Sea Scallop					
1	—	—	146	—	—
2	1-4	5	59	9	25.0
3	7, 8	5	63	10.5	29.5
4	5, 6	5	69	13.5	40.0
5	9-13	5	81	23.0	88.0
6	14, 15	5	75	18.0	50.0
7	7, 8	6	73	17.8	49.0
8	1, 2	6	101	34.5	110
9	3, 6, 9	6	139	78.8	294
11	11	6	130	60.3	235
Bay Scallop					
1	—	—	56	6.4	31.9
2	—	—	56	7.0	33.5
3	4, 10	6	61	10.2	38.1
5	5, 13	6	63	12.5	39.0
6	5, 12	6	59	10.5	33.8

* The length given here denotes the straight-line distance between the middle of the hinge (p) and a point at the margin of the upper shell opposite p. Due to circumstances, the wet meat weight of the sea scallops 2, 3, 4, 5 and 6 was not determined directly but was taken from a graphic representation of data, collected by Mr. J. A. Posgay, relating length and weight of wet meat in over one hundred specimens.

indicated in Figure 2 (animal no. 6, 12 samples). Glass tip or siphon were introduced about 5 mm. into the cloacal chamber. Water from the cloacal cavity was allowed to pass through the siphon (capacity 7 ml.) for about one hour, at a rate of 1.5 ml./min., before the first samples were taken. There were no signs that the presence of tip or siphon, which, as a rule, did not touch the animal's tissues, interfered with the normal vigorous propulsion of water. Slowness of sampling guaranteed that the sample was not contaminated with outside water. A continuous flow of fresh sea water combined with an intensive stirring by means of big bubbles

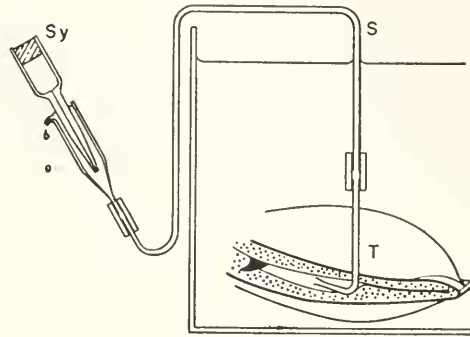


FIGURE 2. Sampling from the exhalant water current in the sea scallop by means of a cloacal siphon. From the continuously overflowing water, samples of about one ml. are taken with a syringe pipette (Sy). The lower end of the glass tube T is about 5 mm. inside the (white) cloacal chamber. Stippled: mantle. Black: tip of gills. Diagram.

of air kept the O_2 -content of the water in the experimental aquarium uniform over prolonged periods of time.

In the earlier studies of the oxygen utilization in Lamellibranchia, made on siphonate forms, it has been assumed that, due to turbulent mixing in the relatively long and narrow siphon, the oxygen in the water leaving the exhalant aperture is fairly uniformly distributed. In the scallops, however, as in many other species, a siphon is lacking and the cloacal chamber communicates with the exterior through a normally widely opened slit. To determine whether appreciable O_2 -gradients occur in the broad ribbon of water escaping from this slit, in the bay scallop two or three samples were taken simultaneously at different spots. The technique used is indicated in Figure 3.

As a rule the introduction of the needles into the cloacal cavity disturbed the ventilation only during the first few minutes and in several cases was accomplished

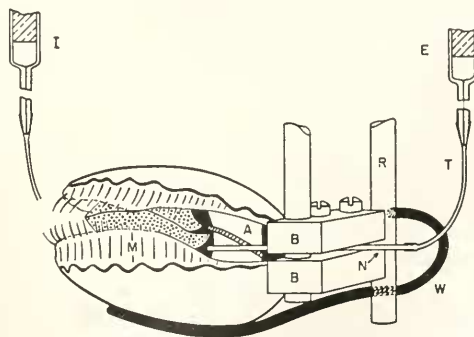


FIGURE 3. Sampling from the exhalant water current in the bay scallop by means of 2 or 3 needles (N), held in a parallel fashion, 3–10 mm. apart, in a plastic block (B). The needles, of which only one is shown, protrude $\frac{1}{2}$ –1 $\frac{1}{2}$ cm. into the cloacal chamber (black), and are connected with a sampling syringe (E). The scallop is paraffined onto a piece of wire (W), held in a plastic rod (R). I: One of the two syringes, sampling simultaneously, at different spots, from inhalant current. M: mantle. Stippled: gills. A: adductor muscle. Hatched: intestine, opening, at the left hand side, into the cloacal chamber (black). Diagram.

without any visible reaction on part of the animal. Moreover, sampling was not begun until after at least one hour had elapsed. On several occasions the needles were left in position overnight. By manipulating two syringes at a time and changing over from one pair to another every 10 seconds, simultaneity of sampling was accomplished to a fair degree. Sampling was interrupted when an animal would interrupt the pumping and was only resumed a couple of minutes after normal ventilation had begun again.

DISCUSSION OF RESULTS

(a) Oxygen consumption in approximately air-saturated water

In Figure 4 the values for the oxygen consumption of the sea and bay scallop studied in the present study (dots and crosses, respectively) are compared with data on the oxygen uptake of various other marine Lamellibranchia selected from the literature. The latter were grouped together and averaged according to geographical distribution (arctic, boreal, and mediterranean) and locomotory faculties (swimming versus non-swimming) of the animals. The relation between these

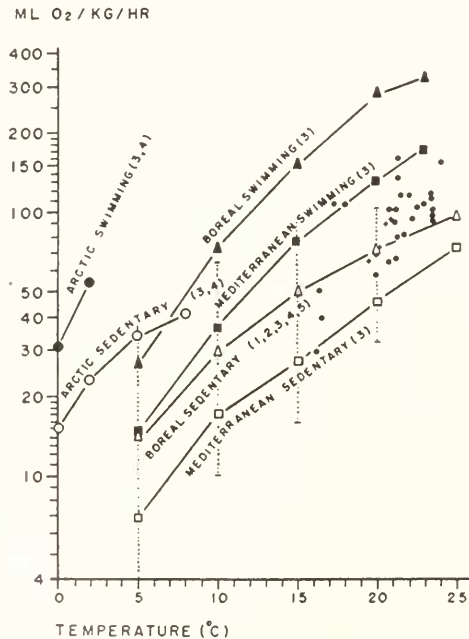


FIGURE 4. Oxygen consumption of resting specimens of bay and sea scallop studied in the present paper (crosses and dots, respectively) compared with the oxygen uptake of other marine Lamellibranchia, selected from the literature and grouped together according to geographical distribution (arctic, boreal and mediterranean species) and according to locomotory faculties (swimming or more or less sedentary species). On the curves, the points represent the arithmetical mean of all Q_{O_2} -values available for a given temperature. The figures in brackets refer to the source in the literature from which the data, used to compute these mean values, were taken, as follows: 1 = Bruce, 1926; 2 = Thamdrup, 1935; 3 = Spärck, 1936; 4 = Thorson, 1936; 5 = Wheldon and Sommer, 1937. Vertical, stippled, lines: spread of data in the group of the boreal sedentary species.

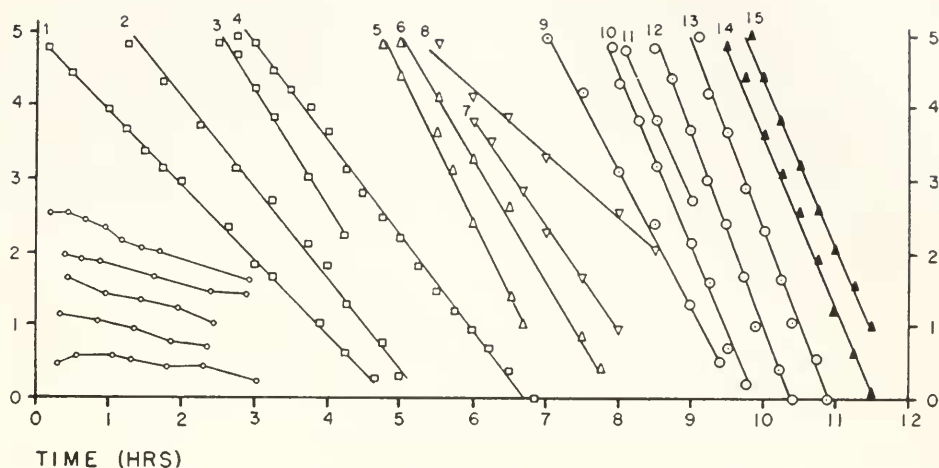
ML O₂ IN SYRINGE

FIGURE 5. Rate of oxygen uptake in the sea scallop in approximately air-saturated water. Ordinate: amount of oxygen (ml.) in the measuring syringe. Abscissa: time (hrs.). Numbering of tests as in Table I. Small circles: blank runs.

factors and the metabolic rate in marine Lamellibranchia has been dealt with already by Spärck (1936). The following data (adult specimens, outside the spawning season) were used: Spärck (1936): 9 arctic, 12 boreal, and 11 mediterranean species; Thorson (1936): 11 arctic species; Thamdrup (1935): 3 boreal species; Bruce (1926): one boreal species; and Whedon and Sommer (1937): one boreal species. The figure indicates that the O₂-consumption in the bay scallop is essentially the same as that in the sea scallop. Over the range of temperatures investigated (16–24° C.), the O₂-uptake in these species is somewhat intermediate in position between the O₂-uptake of mediterranean scallops and that of more sedentary bivalves from that area and thus is about the same as the O₂-uptake of the average boreal non-swimming lamellibranch. In using the curves in Figure 4 as a basis for comparison, it must be borne in mind, however, that they give a greatly simplified picture inasmuch as the data, from which they were computed, show a very considerable spread in most cases. The spread in the Q_{O₂}-values of the different species of lamellibranchs in the boreal-sedentary group, is indicated in the figure (stippled lines). Not enough data are available to establish, in the bay and sea scallop, a relation between Q_{O₂} and season or size, or to estimate Q₁₀.

For several species of bivalve molluscs, lengthy periods of stoppage of the water propulsion have been reported in the literature. To the list of species given by van Dam (1938, p. 123; cf. also Verwey 1952, pp. 189–193) can be added *Venus mercenaria* (Collip, 1921; Chipman and Galtsoff, 1949) and *Hyridella australis* (Hiscock, 1950; 1953). Persistence of rhythmicity in O₂-uptake in molluscs under laboratory conditions was further reported by Gompel (1937) and by Sandeen *et al.* (1953), and in the rate of water propulsion in the California mussel by Rao (1953). No prolonged interruptions of the ventilation were observed in the scallops and the

oxygen uptake of the latter, accordingly, appears to be quite uniform (Figs. 5 and 6). Also, as already mentioned by Spärck (1936), scallops cannot be kept out of water for nearly as long a time as many other bivalves, without being damaged. When kept in moist air at room temperature they soon open their valves and gradually lose the typical reaction to the sudden cast of a shadow which consists of a vigorous closing movement. The oxygen consumption determined in a sea scallop which had been subjected to such a treatment for seven hours appeared to be low for the temperature of the test (74 ml. O_2 /kg./hr., 22.7° C.), the critical oxygen tension high (test 3, Fig. 6). During this test the animal held its valves abnormally wide apart and its gills were arranged in a disorderly fashion.

(b) *Oxygen consumption at progressively decreasing concentrations of oxygen*

With not too rapid a fall in the oxygen content of the inspired water, the scallops appeared to be capable of maintaining a normal oxygen uptake down to a concentration of oxygen of about 1.0–0.5 ml. O_2 /L. (Fig. 6). Similar findings have been

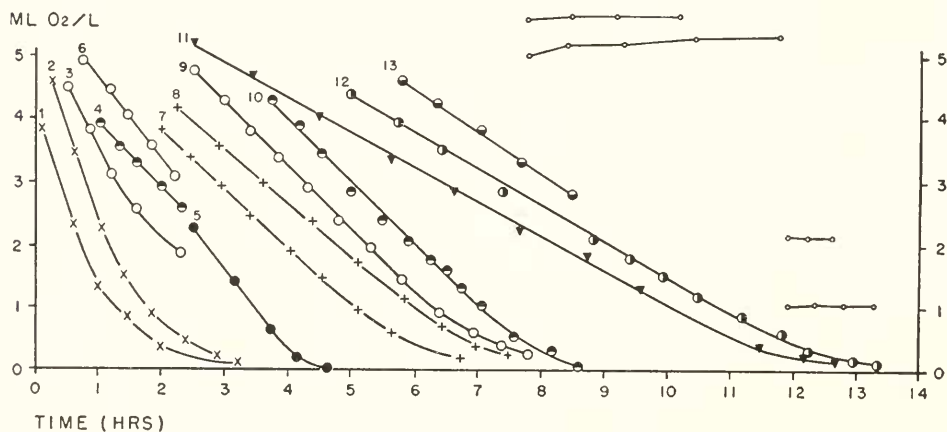


FIGURE 6. Rate of oxygen uptake of the sea scallop and of the bay scallop at progressively decreasing tensions of oxygen. Ordinate: O_2 -content of water in respiratory chamber (ml. O_2 /L.). Abscissa: time (hrs.). Numbering of tests as in Table I. For discussion of test no. 3, see text. Small circles: blank runs.

reported for other species of Lamellibranchiata (Weiland, 1919; Nozawa, 1929; Galtsoff and Whipple, 1930; Ishida, 1935; Thamdrup, 1935; Whedon and Sommer, 1937; Hers, 1943). To what extent this faculty is based on an increase in the amount of water pumped or on an increase in the percentage oxygen utilization, has not been investigated. It was noticed that the animals, in water poor in oxygen, display little or no active motion, gradually lose the "shadow-cast reaction," and erect the pallial velum in a vertical fashion. It is possible that the vigorous stirring of the water in the respiration chamber has contributed to some extent to the ventilation of the animal's tissues with water and thus has lowered the critical oxygen tension below a level which would prevail if this ventilation depended solely on the currents generated by the animal itself.

(c) Oxygen utilization

In the sea scallops the oxygen utilization was found to range from about $\frac{1}{2}$ to 9% ; in four out of the five bay scallops investigated the values represent a range of about 1 to 13% (Table II). Similarly low values have been found in siphonate Lamellibranchia and in several other filter feeding animals (van Dam, 1935b, 1938; Hazelhoff, 1938). Low values are also obtained for the oxygen utilization calculated from data on oxygen consumption and rate of water propulsion as given for a number of filter feeders, e.g., by Jørgensen (1952). It seems, then, that true filter feeders normally pump large amounts of water (*cf.* also Loosanoff and Nomejko, 1946; Owen, 1953), from which only a small fraction of the dissolved oxygen is being consumed. It would be of interest to determine the oxygen utilization or the rate of water propulsion in those species of bivalves which are not true filter

TABLE II
Percentage oxygen utilization in the sea scallop*
O₂-content of inhaled water about 5 ml. O₂/L. Temperature about 17–22° C.

Specimen no. 1	Specimen no. 5	Specimen no. 6
2.9	7.5	1.9
2.2	6.4	2.7
0.7	5.2	0.8
1.6	7.6	3.6
1.6		2.3
1.6	Mean: 6.6%	2.3
4.0		2.9
8.8		3.4
1.8		1.9
4.0		3.8
2.2		3.8
2.2		3.8
0.4		
1.0		Mean: 2.8%
Mean: 2.5%		

* Except for one (Specimen no. 4, utilization 0.9%), the utilization values obtained in the bay scallop are shown in Figure 7.

feeders as, for example, those feeding on bottom deposits (*cf.* Yonge, 1953).

In one of the bay scallops which ventilated much less vigorously than any of the other specimens investigated, high percentage oxygen utilization values (13–72%) were found. A similar, slow, often interrupted, ventilation entailing a high oxygen utilization was observed in some individuals of Anodonta (van Dam, 1938). In the specimens of Anodonta investigated by Koch and Hers (1943; personal communication) a high utilization prevailed throughout. Whether a slow rate of ventilation, as displayed by these specimens of Pecten and Anodonta, can be considered normal, *i.e.*, will provide the animal with enough oxygen and food to sustain normal, aerobic, respiration and growth, is not known.

In the stream of water escaping from the cloacal chamber of the bay scallops considerable gradients in the concentration of oxygen occur. These gradients are

shown, in Figure 7, in the form of utilization values obtained simultaneously, but at different spots, in the exhalant water current of one and the same specimen. These values were connected by a line parallel to the ordinates. The steepest oxygen concentration gradients occurred in those cases where the utilization was high. In one of these cases (the group of three dots closest to the right hand ordinate) the utilization values were 33.4%, 37.4%, and 71.5%, respectively, representing oxygen concentrations of 66.6%, 62.6%, and 28.5%, of the oxygen content of the inhaled water. In this case, then, the concentration of oxygen at one spot was more than twice as high as at another spot. In the other, more vigorously ventilating specimens, in which the utilization was much lower, the differences in oxygen concentration did not exceed 10% of the O_2 -content of the inhaled water.

Inhomogeneity in the distribution of oxygen in the exhalant water current as observed in the bay scallop by the present author and in *Anodonta* by H. J. Koch

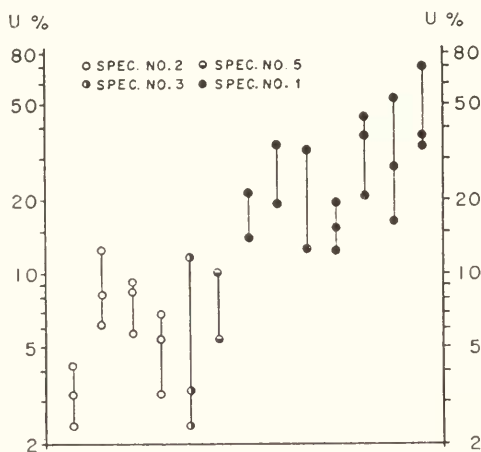


FIGURE 7. Gradients in the concentration of oxygen in the exhalant water current in the bay scallop, presented as the percentage oxygen depletion ($U\%$) of samples drawn simultaneously at different spots in one and the same animal. $U\%$ -values from simultaneously drawn samples are connected by a line parallel to the ordinates. O_2 -content inhaled water approx. 4.5–5 ml. O_2/L ; temperature about 20–24.5° C.

and M. J. Hers (personal communication), of course, excludes the possibility of determining the true average oxygen utilization with a sampling method. Only if all the water leaving the animal could be collected would the determination of such a value be possible. The oxygen utilization values, then, obtained in this and in previous investigations, are only approximations of this average value.

I wish to thank Dr. A. C. Redfield and Dr. P. F. Scholander for stimulating interest and valuable criticism, and Mr. J. A. Posgay for help in the procurement of scallops.

SUMMARY

1. The oxygen uptake and the percentage of oxygen withdrawn from the inhaled water, were determined in resting specimens of two species of lamellibranch mol-

luses which have a capacity for rapid swimming locomotion, *viz.* in the sea scallop (*Pecten grandis* Sol.) and in the bay scallop (*Pecten irradians* Lam.).

2. In accordance with the absence of prolonged ventilation pauses common in several other species of Lamellibranchia, the oxygen uptake in the bay and sea scallop is quite uniform. In both species Q_{O_2} was found to be about 70 ml./kg./hr. at 20° C. This value is about half the Q_{O_2} -value found in swimming species from the mediterranean and falls well within the range of Q_{O_2} -values in boreal non-swimming types of lamellibranchs recorded in the literature.

3. The oxygen uptake was independent of the oxygen tension down to a concentration of oxygen of about 1-½ ml. O_2 /L.

4. The percentage of oxygen withdrawn from the inhaled water, in most cases, was low, ranging, approximately, from ½ to 13%. It was demonstrated that in the stream of water escaping from the cloacal chamber of the bay scallop considerable gradients in the concentration of oxygen occur.

LITERATURE CITED

- BRUCE, J. R., 1926. The respiratory exchange of the mussel (*Mytilus edulis* L.). *Bioch. J.*, **20**: 829-846.
- CHIPMAN, W. A., AND P. S. GALTISOFF, 1949. Effects of oil mixed with carbonized sand on aquatic animals. *U. S. Dep. Int. Fish and Wildlife Serv. Spec. Sc. Rep. Fish.*, **1**: 1-52.
- COLLIP, J. B., 1921. A further study of the respiratory processes in *Mya arenaria* and other marine mollusca. *J. Biol. Chem.*, **49**: 297-310.
- GALTISOFF, P. S., AND D. V. WHIPPLE, 1930. Oxygen consumption of normal and green oysters. *Bull. U. S. Bur. Fish.*, **46**: 489-508.
- GOMPEL, M., 1937. Recherches sur la consommation d'oxygène de quelques animaux aquatiques littoraux. *C. R. Acad. Sci.*, **205**: 816-818.
- HAZELHOFF, E. H., 1938. Ueber die Ausnutzung des Sauerstoffs bei verschiedenen Wassertieren. *Zeitschr. vergl. Physiol.*, **26**: 300-327.
- HERS, M. J., 1943. Relation entre respiration et circulation chez *Anodonta cygnea* L. *Ann. Soc. Roy. Zool. Belgique*, **74**: 45-54.
- HISCOCK, I. D., 1950. Shell movements of the freshwater mussel *Hyridella australis* Lam. (Lamellibranchiata). *Austr. J. Mar. Freshwater Res.*, **1**: 259-268.
- HISCOCK, I. D., 1953. Osmoregulation in Australian freshwater mussels (Lamellibranchiata). II. Respiration and its relation to osmoregulation in *Hyridella australis* (Lam.). *Austr. J. Mar. Freshwater Res.*, **4**: 330-342.
- ISHIDA, S., 1935. On the oxygen consumption in the oyster *Ostrea gigas* Thunberg under various conditions. *Sci. Rep. Tôhoku Univ., Ser. 4*, **10**: 619-638.
- JØRGENSEN, C. B., 1952. On the relation between water transport and food requirements in some marine filter feeding invertebrates. *Biol. Bull.*, **103**: 356-363.
- KOCH, H. J., AND M. J. HERS, 1943. Influence de facteurs respiratoires sur les interruptions de la ventilation par le siphon exhalant chez *Anodonta cygnea* L. *Ann. Soc. Roy. Zool. Belgique*, **74**: 32-44.
- LOOSANOFF, V. L., AND C. A. NOMEJKO, 1946. Feeding of oysters in relation to tidal stages and to periods of light and darkness. *Biol. Bull.*, **90**: 244-264.
- NOZAWA, A., 1929. The normal and abnormal respiration in the oyster *Ostrea circumpecta* Pils. *Science Rep. Tôhoku Imp. Univ., Ser. 4*, **4**: 259-325.
- OWEN, G., 1953. On the biology of *Glossus humanus* (L.) (Isocardia Cor Lam.). *J. Mar. Biol. Assoc.*, **32**: 85-106.
- RAO, K. P., 1953. Rate of water propulsion in *Mytilus californianus* as a function of latitude. *Biol. Bull.*, **104**: 171-181.
- SANDEEN, M. I., G. C. STEPHENS AND F. A. BROWN, JR., 1953. Persistent diurnal rhythms of O_2 -consumption in the periwinkle, *Littorina littorea*, and the oyster drill, *Urosalpinx cinereus*. *Biol. Bull.*, **105**: 381-382.

- SCHOLANDER, P. F., 1949. Volumetric respirometer for aquatic animals. *Rev. Sci. Instr.*, **20**: 885-887.
- SPÄRCK, R., 1936. On the relation between metabolism and temperature in some marine lamellibranchs, and its zoogeographical significance. *K. Danske Vid. Selsk. Biol. Medd.*, **13** (5): 1-27.
- THAMDRUP, H. M., 1935. Beiträge zur Ökologie der Wattenfauna auf experimenteller Grundlage. *Medd. Komm. Danmarks Fiskeri og Havunders, Ser. Fiskeri*, **10**: 2.
- THORSON, G., 1936. The larval development, growth, and metabolism of arctic marine bottom invertebrates compared with those of other seas. *Medd. om Gronland*, **100**: 1-155.
- VAN DAM, L., 1935a. A method for determining the amount of oxygen dissolved in 1 cc. of water. *J. Exp. Biol.*, **12**: 80-85.
- VAN DAM, L., 1935b. On the utilization of oxygen by *Mya arenaria*. *J. Exp. Biol.*, **12**: 86-94.
- VAN DAM, L., 1938. On the utilization of oxygen and regulation of breathing in some aquatic animals. Thesis, Groningen.
- VERWEY, J., 1952. On the ecology of distribution of cockle and mussel in the Dutch Waddensea. Their role in sedimentation and the source of their food supply. With a short review of the feeding behaviour of bivalve molluscs. *Arch. Néerl. Zool.*, **X**: 171-239.
- WEINLAND, E., 1919. Beobachtungen ueber den Gaswechsel von *Anodonta cygnea* L. *Zeitschr. Biol.*, **69**: 1-86.
- WHEDON, W. F., AND H. SOMMER, 1937. Respiratory exchange of *Mytilus californianus*. *Zeitschr. vergl. Physiol.*, **25**: 523-528.
- YONGE, C. M., 1953. Mantle chamber and water circulation in the Tridacnidae (Mollusca). *Proc. Zool. Soc. London*, **123**: 551-561.